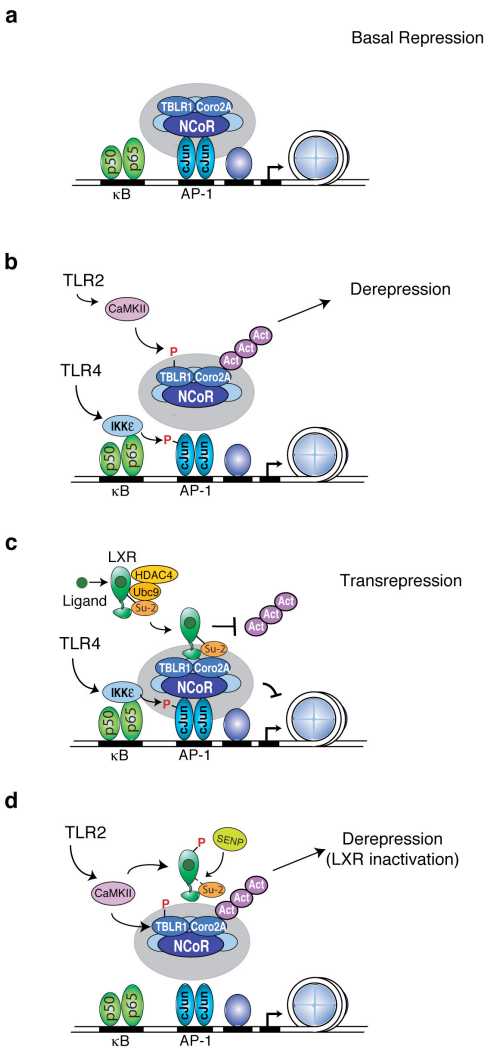
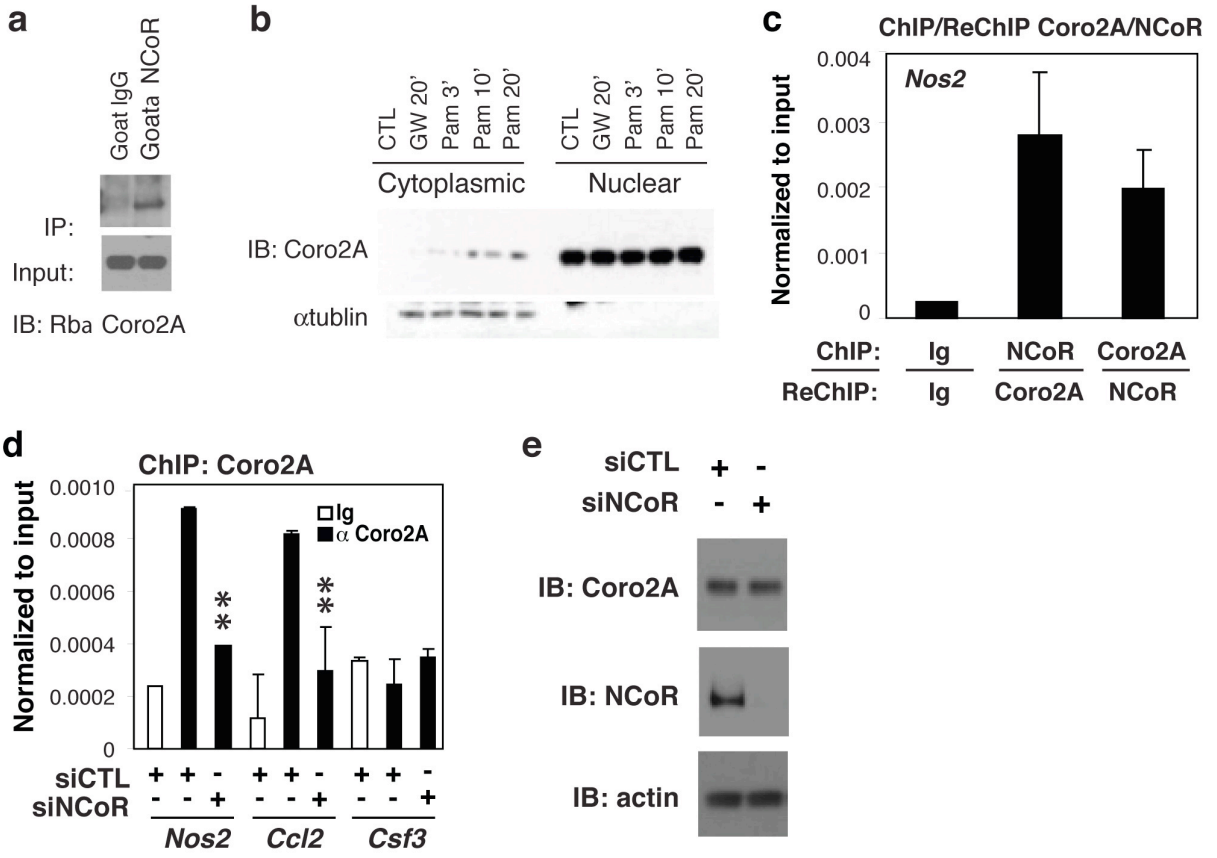
## Supplementary Figures



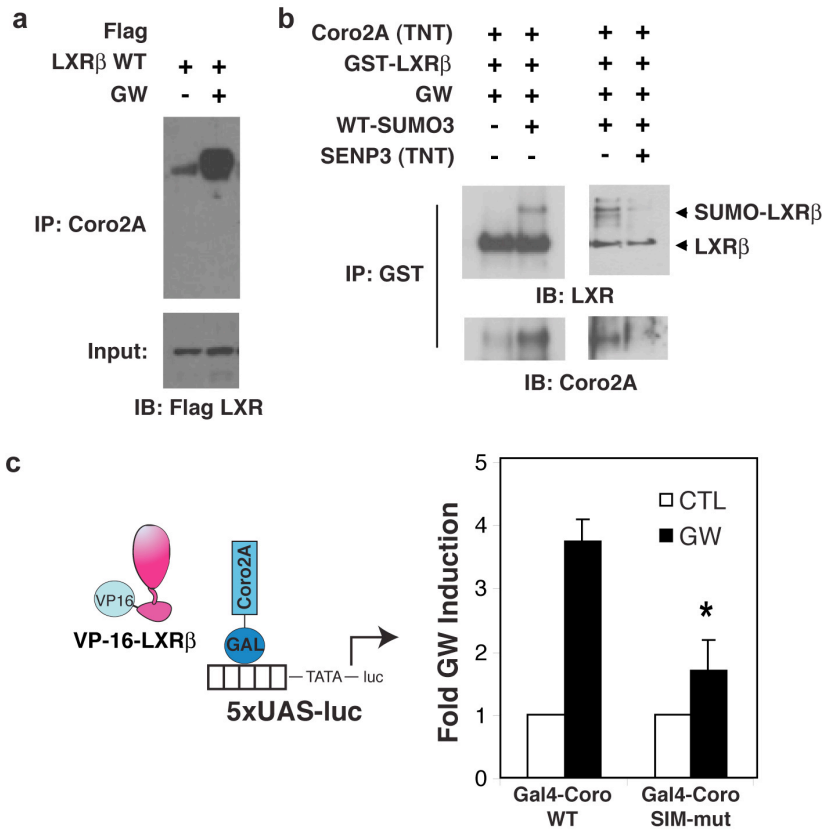

**Supplementary Figure 1. Working model for roles of Coro2A, CaMKIIγ and SENP3 in NCoR clearance and LXR transrepression.**

**a**, In resting macrophages, NCoR complexes reside on inflammatory responsive promoters through interactions with non-phosphorylated cJun and mediate basal repression. **b**, TLR2 and TLR4 signaling initiate recruitment of oligomeric actin to the Coro2A component of the NCoR complex, which is required for dissociation of the NCoR complex from the promoter. TLR2 signaling initiates NCoR clearance through CaMKII-induced phosphorylation of TBLR1. TLR4 signaling initiates NCoR clearance through IKKε-induced phosphorylation of cJun. **c**, Ligand dependent SUMOylation of LXRs result in their docking to Coro2A. This prevents signal-dependent interaction between Coro2A and oligomeric actin in response to LPS. As a consequence, NCoR complexes remain bound and continue to exert a repression function. **d**, Signaling through TLR2 results in CaMKII-dependent phosphorylation of LXR, producing a docking site for SENP3. SENP3 cleaves SUMO from LXR, thereby inactivating its transrepression function.

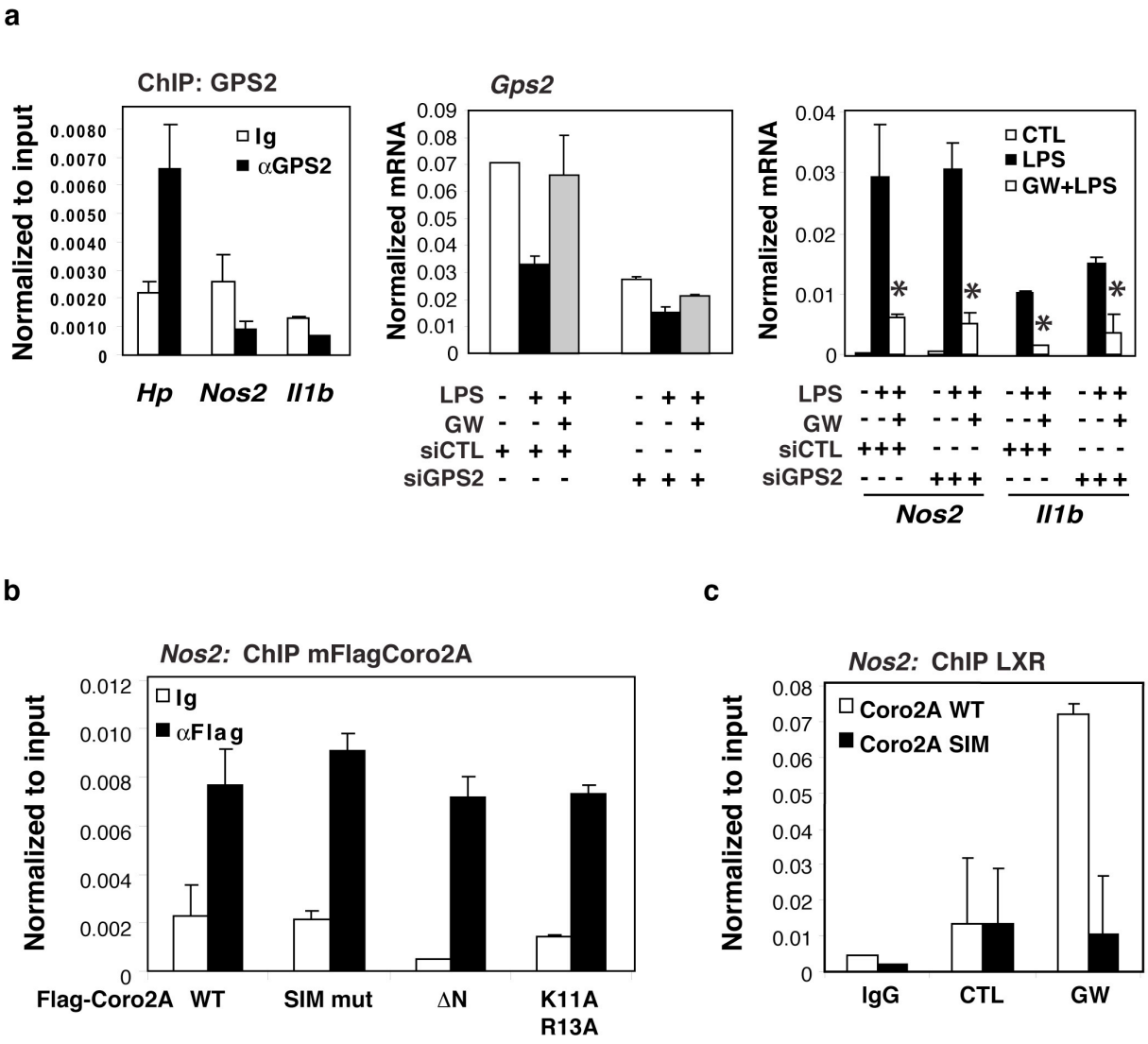



**Supplementary Figure 2. Subcellular localization of Coro2A and interaction with NCoR.**

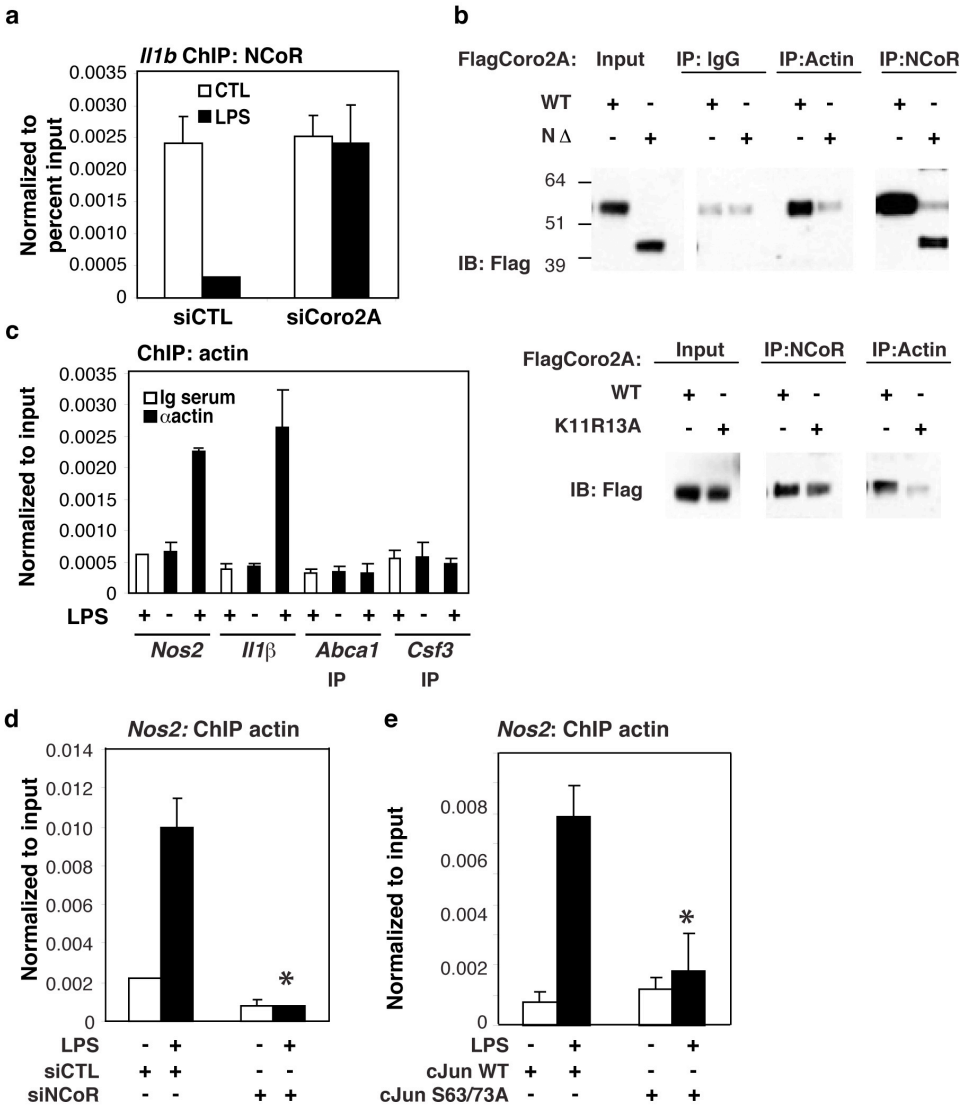
**a,** Whole cell extracts from BMDM were subjected to immunoprecipitation using control or NCoR specific antibodies. Precipitates were resolved by SDS-PAGE and analyzed by IB with Coro2A antibodies. **b,** Nuclear and cytoplasmic protein extracts from BMDM treated with 1μM GW3965 or 300ng/ml Pam3CSK4 were immunoblotted for Coro2A and α-tubulin. **c**, ChIP/Re-ChIP for Coro2A-NCoR binding to the *Nos2* promoter in BMDMs. **d,** ChIP for Coro2A-binding to *Nos2*, *Ccl2* and *Csf3* promoters in BMDMs after siRNA transfection. **e**, Immunoblotting of Coro2A, NCoR and actin in BMDMs transfected with control or NCoR-specific siRNAs for 48h.




**Supplementary Figure 3. Interaction of Coro2A with SUMOylated LXRs.**
**a**,HeLa cells were transfected with an expression vector for Flag-LXRβ and treated with GW3965 (GW) or control solvent for 1h. Whole cell lysates were subjected to IP with Coro2A antibody and precipitates were immunoblotted for Flag. **b,** GST-LXRβ was SUMOylated in vitro with SUMO3, and de-SUMOylated by incubation with in vitro transcribed and translated SENP3. GST-LXRβ, SUMO3-GST-LXRβ and de-SUMOylated SUMO-GST-LXRβ were captured on a glutathione affinity matrix and incubated with in vitro-translated (TNT) Coro2A. Following washing, captured LXR and interacting Coro2A were detected by immunoblotting using LXR and Coro2A-specific antibodies. **c**, RAW264.7 cells were transfected with a 5xUAS-luciferase reporter gene, and plasmids directing expression of VP16-LXRβ and GAL4 DNA binding domain fused to wild type (WT) Coro2A or Coro2A with a SIM domain mutation (SIM-mut). Cells were treated with GW for 16h and luciferase activity measured. Data is plotted as average fold induction by GW ± s.e.m. *P<0.05 for fold induction of SIM-mut vs WT.

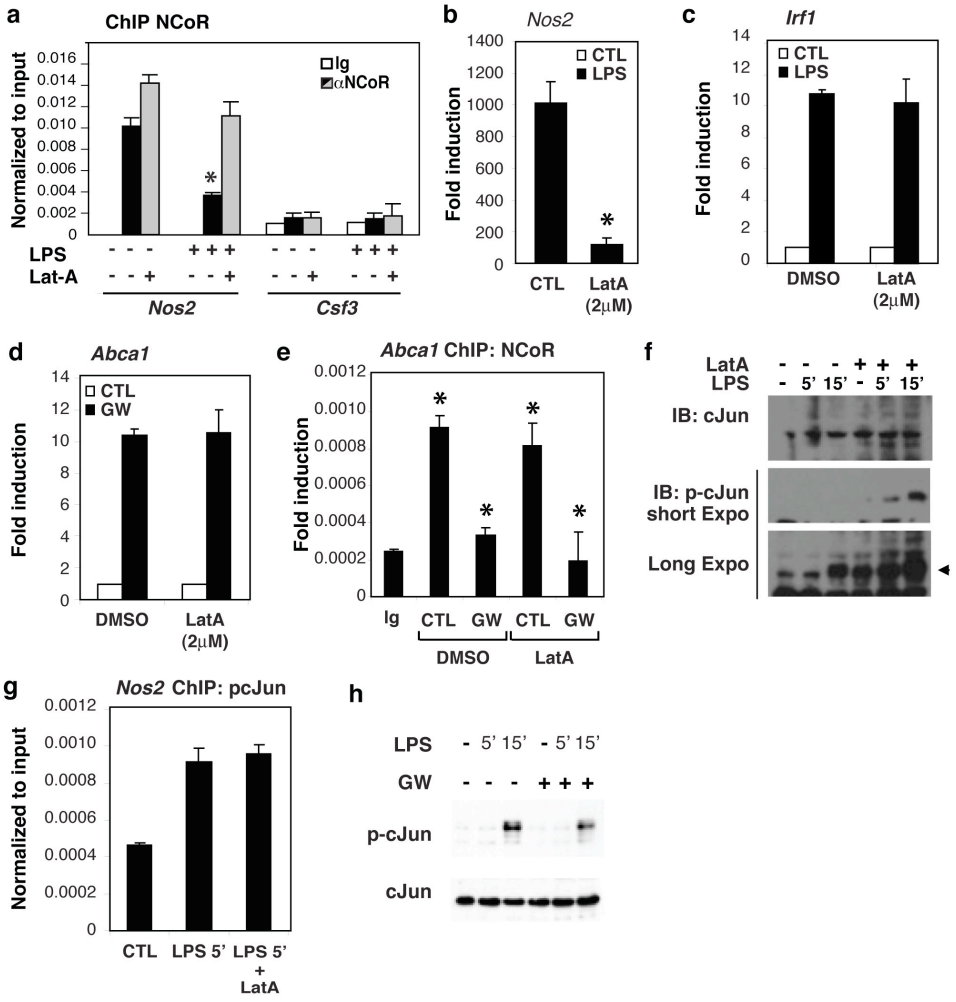



**Supplementary Figure 4. Evaluation of GPS2 and mutant forms of Coro2A.** **a,** (Left) ChIP for GPS2-binding to the *Hp, Nos2* and *Il1b* promoters in BMDM. (Middle) RNA was extracted from BMDMs transfected with control or *Gps2*-specific siRNAs for 48 hrs. *Gps2* mRNA levels were determined by Q-PCR and normalized to *Gapdh*. (Right) BMDMs were transfected with control or GPS2 specific siRNAs for 48 hrs. Cells were pretreated with or without synthetic LXR ligand (1μM, GW3965), followed by 100ng/ml LPS for 6hrs. *Nos2*, *Il1b* and *Gps2* mRNA was quantified by Q-PCR and normalized to *Gapdh*. **b**, RAW264.7 cells were transfected with the indicated Flag-Coro2A expression plasmids and the *Nos2*-luciferase reporter gene. ChIP was performed using anti-Flag antibody and *Nos2*-promoter sequences were quantified by Q-PCR. **c,** RAW264.7 cells were transfected with the indicated Flag-Coro2A expression plasmids and the *Nos2*-luciferase reporter gene. ChIP was performed using anti-LXR antibody and *Nos2*-promoter sequences were quantified by Q-PCR. (In all cases, data is plotted as averages ± s.e.m., *P<0.05 GW treated WT versus SIM mutant).

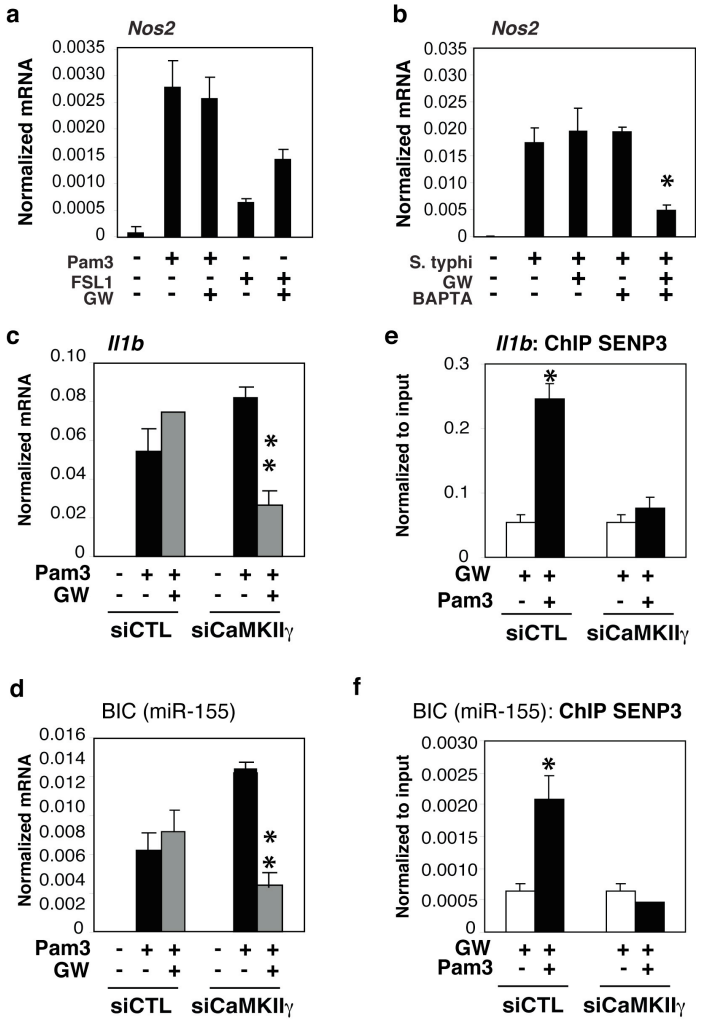



**Supplementary Figure 5. Evaluation of interactions of WT and mutant forms of Coro2A with NCoR and actin.**

**a,** BMDMs were transfected with control or Coro2A-specific siRNAs for 48 hours. Cells were treated with vehicle or LPS for 1h and ChIP was performed for the *Il1b* promoter using an NCoR-specific antibody. **b,** HeLa cells were transfected with expression vector for either WT or mutants of Flag-Coro2A as indicated. 48hrs after transfection, IPs were carried out with control, actin, or NCoR-specific antibodies and analyzed by IB with Flag antibodies. **c.** ChIP for actin-binding to the *Nos2*, *Il1b*, *Abca1* and *Csf3* promoters in BMDMs treated with LPS. **d,** ChIP for actin on the *Nos2* promoter in BMDM 48hrs after transfection of control scrambled or NCoR specific siRNA and treated with 100ng/ml LPS for 5 mins. **e,** ChIP for actin on the *Nos2*-luc promoter in RAW264.7 cells transfected with *Nos2*-luc reporter plasmids and with expression vectors for c-Jun wild-type or phosphor-null mutant (S63/73A) as indicated and treated with LPS for 5 mins.

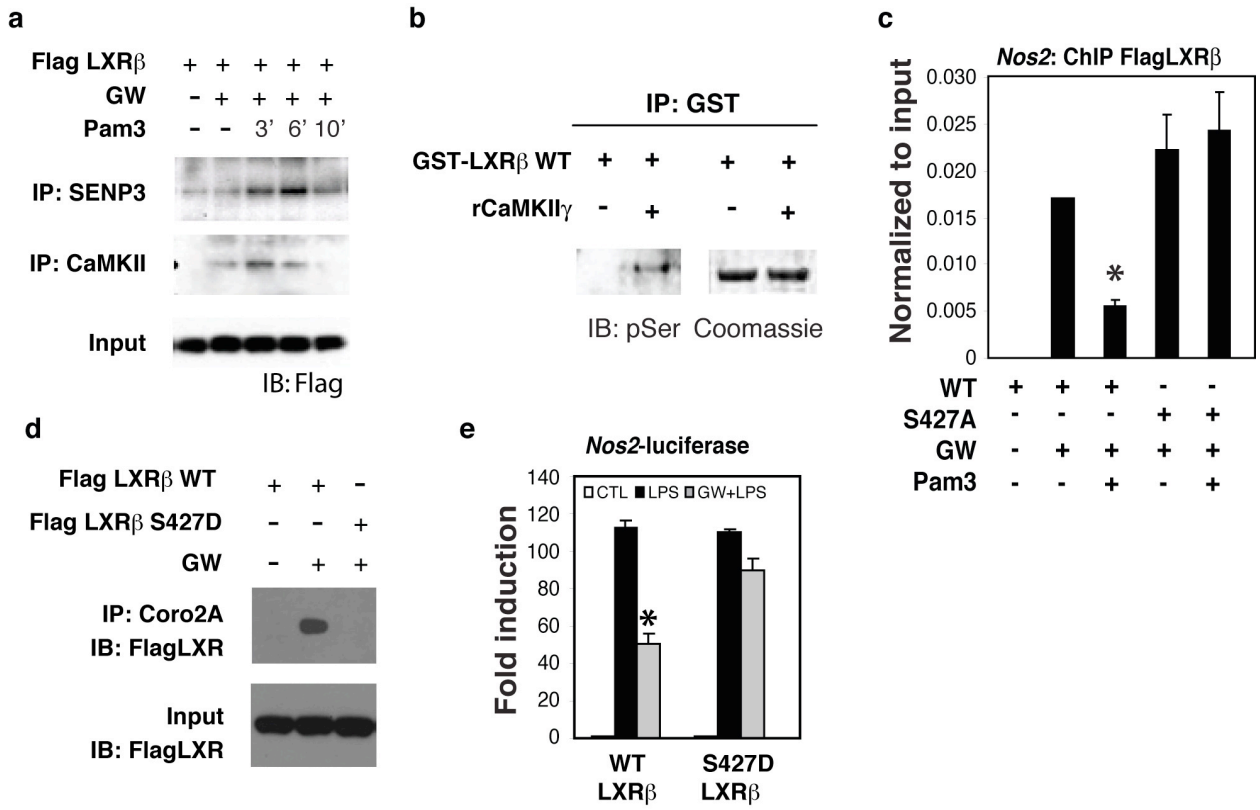



**Supplementary Figure 6. Effects of short-term treatment with latrunculin A.** **a.** ChIP for NCoR-binding to the *Nos2* promoter in BMDMs treated with LPS in the presence or absence of latrunculin A (2μM). **b-c,** BMDMs were treated with vehicle or LPS for 6h in the presence or absence of latrunculin A (LatA). *Nos2* and *Irf1* mRNA levels were quantified by Q-PCR and fold induction in response to LPS was calculated. **d**, BMDMs were treated with vehicle or GW3965 for 6h in the presence or absence of latrunculin A (LatA). *Abca1* mRNA levels were quantified by Q-PCR and fold induction in response to GW3965 was calculated. **e**, BMDMs were treated with vehicle or GW3965 for 1h in the presence or absence of latrunculin A (LatA). ChIP was performed for NCoR on the *Abca1* promoter. (*P<0.04 CTL versus GW treatment). (Averages ± s.e.m., *P<0.04 versus siCTL or cJun WT LPS). **f,** IB of cJun and phospho-(p)-cJun in BMDMs pretreated with or without 2μM latrunculin A (LatA) and stimulated with 100ug/ml LPS for the indicated times. **g**, BMDMs were treated with vehicle or LPS for 5 min in the presence or absence of latrunculin A (LatA). ChIP was performed for phospho-cJun on the *Nos2* promoter. (*P<0.04 LPS versus CTL treatment). **h**, IB of cJun and phosphocJun in BMDM pretreated with or without 1μM GW3965 and stimulated with 100ug/ml LPS for the indicated times.

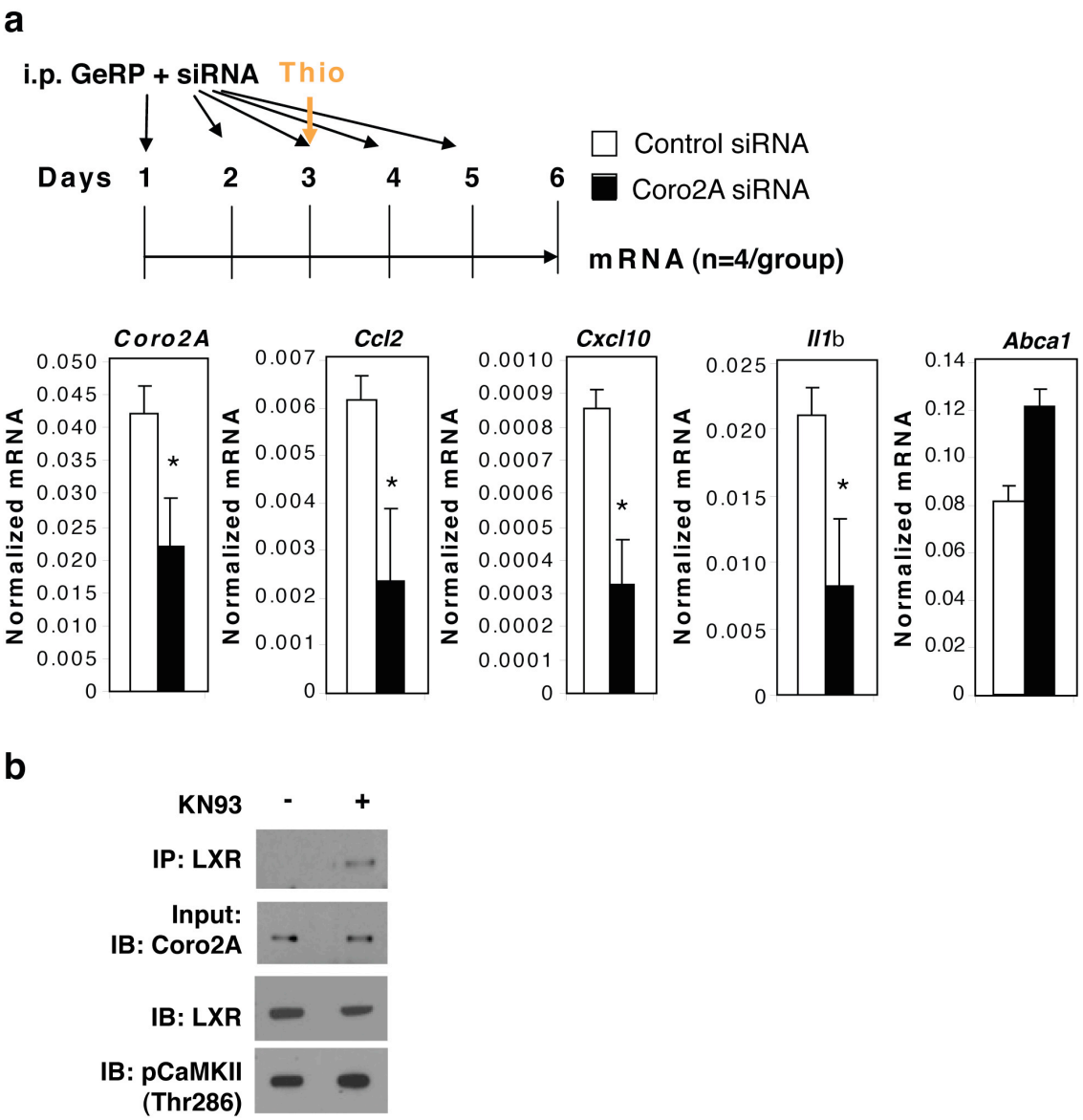



**Supplementary Figure 7. Evaluation of TLR1/2 signaling and CaMKII on LXR transrepression.**

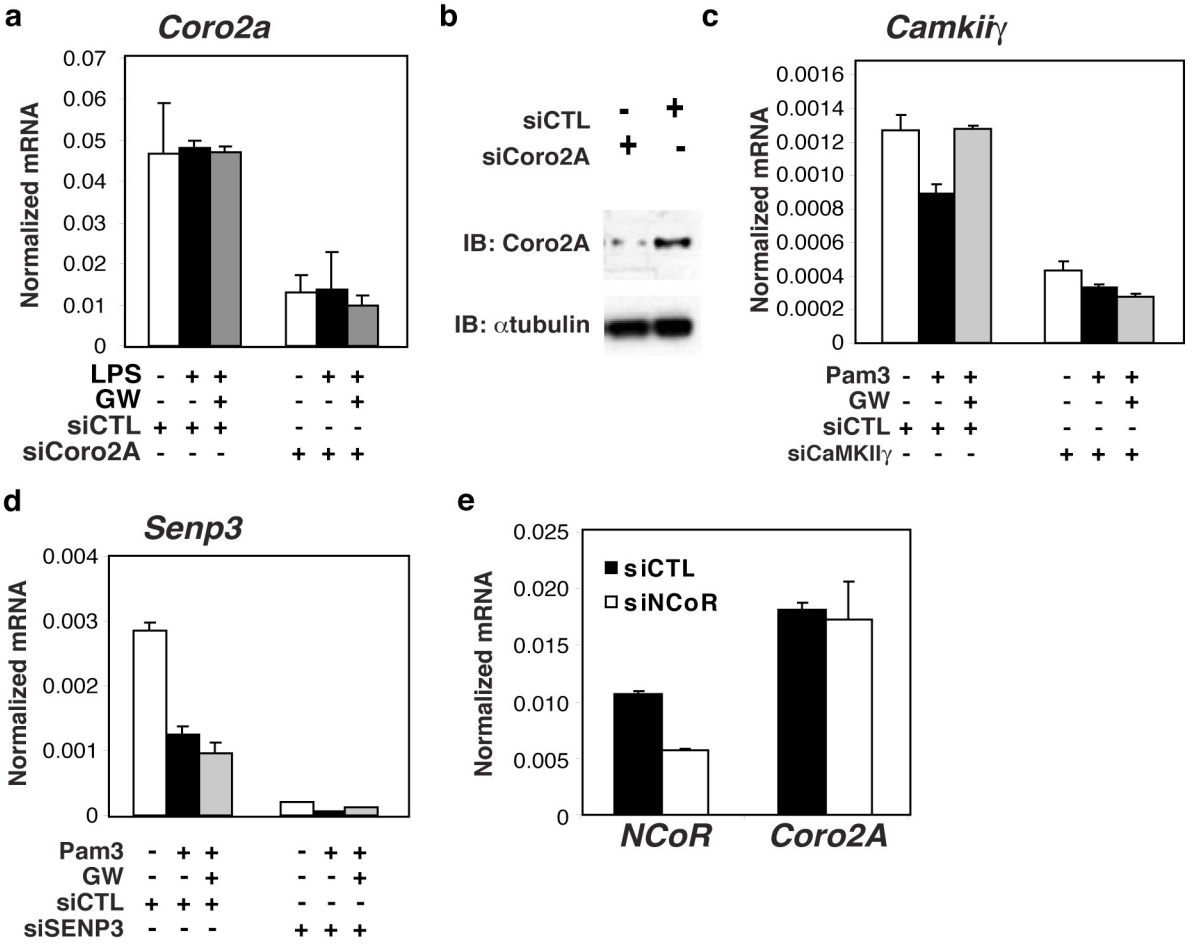
**a**, BMDMs were treated with GW3965 and then the TLR2/6 agonist (FSL-1) or the TLR1/2 (Pam3) agonist for 6h, as indicated. *Nos2* mRNA was quantified by Q-PCR and normalized to *Gapdh*. **b**, BMDMs were treated with GW3965 and incubated with *Salmonella typhimurium* in the presence or absence of the calcium chelating agent BAPTA for 6h, as indicated. *Nos2* mRNA was quantified by Q-PCR and normalized to *Gapdh* (Averages ± s.e.m., *P<0.04 BAPTA versus non-treated cells). **c-d**, BMDMs were transfected with control or CaMKIIγ-specific siRNAs for 48 hours. Cells were treated with vehicle or Pam3 in the presence of GW3965 as indicated for 6h and *Il1b* or *BIC* (miR155) mRNA were quantified by Q-PCR and normalized to *Gapdh*. (Averages ± s.e.m., *P<0.04 versus Pam3 alone). **e-f**, ChIP for LXR and NCoR-binding to the *Il1b* and *BIC* promoter in BMDM 48hrs after transfection of control scrambled or SENP3 specific siRNA. Cells were pretreated with or without 1μM GW3965 and then stimulated with 300ng/ml Pam3CSK for 1hr. (Averages ± s.e.m., **P<0.01 versus Pam3, *P<0.02 versus control).




**Supplementary Figure 8. Phosphorylation of LXRβ and interaction with SENP3.** **a,** RAW cells were transfected with Flag-LXRβ expression plasmid and treated with GW3965 and Pam3 as indicated. Whole cell lysates were subjected to immunoprecipitation using anti-SENP3 or anti-CaMKIIg antibodies and immunoprecipitates analyzed by immunoblotting with anti-Flag antibody. **b**, Full length rGST-hLXRβ IP on glutathione agarose was incubated with or without 0.5μg of activated rCaMKIIγ as indicated. GST-LXR was recovered and resolved by SDS-PAGE and immunoblotted for phosphoserine. **c,** ChIP for Flag-LXR on the *Nos2*-luc promoter in RAW264.7 cells transfected with *Nos2*-luc reporter plasmids and wild-type or mutant Flag-LXRβ as indicated, and treated with 1μM of GW3965 for 1hr and challenged with 300ng/ml Pam3CSK4 for 1hr. (Averages ± s.e.m., *P<0.01 versus WT GW). **d,** RAW264.7 cells were transfected with WT or mutant FlagLXRβ in the presence or absence of GW3965. Whole-cell lysates were subjected to IP with Coro2A specific antibodies and detected by anti-Flag antibody on IB. **e**, RAW264.7 cells were transfected with the *Nos2*-luciferase reporter gene and expression plasmids for WT LXRβ or LXRβ S427D. Cells were treated with LPS in the presence or absence of GW3965 for 6h as indicated. Luciferase activity was measured and fold induction to LPS-treatment was calculated. (Averages ± s.e.m., *P<0.02 versus LPS alone).




**Supplementary Figure 9. Effects of Coro2A knockdown in vivo using GeRP delivery. a.** GeRP delivery spheres were loaded with control or Coro2A-specific siRNAs and injected into the peritoneal cavities of mice according to the indicated schedule (n=4 per condition). Thioglycollate was injected on day 3 and intraperitoneal macrophages were recovered on day 6 and assayed for expression of the indicated genes by Q-PCR. Values were normalized to Gapdh. (n=4 per condition). (Averages ± s.e.m., *P<0.05 versus siCTL treated animals). Statistical significance was determined by a two-tailed Student's t-test. **b,** Coimmunoprecipitation assay probing for the interaction between endogenous Coro2A and LXR in peritoneal exudates cells from mice challenged with thioglycollate medium and treated i.p. with CaMKII inhibitor, KN93 or vehicle. Lysates were subject to IP with LXR antibodies and analyzed by IB against Coro2A. Activation of CaMKII was determined by IB for phosphorylation of Thr286.




**Supplementary Figure 10. Efficacy of Coro2A, CaMKIIg and Senp3 knockdown**
**a**, BMDMs were transfected with control or Coro2A-specific siRNAs for 48hrs and pretreated with or without 1μM GW3965, followed by 6hrs of treatment with 100ng/ml LPS as indicted. *Coro2a* mRNA levels were determined by Q-PCR and normalized to *Gapdh*. **b**, BMDMs were transfected with control or Coro2A-specific siRNAs for 48hrs. Whole cell lystates were resolved by SDS-PAGE and immunoblotted for Coro2A. **c-e**, BMDMs were transfected with control, CamkIIg–, Senp3-, or NCoR specific siRNAs for 48hrs and pretreated with or without 1μM GW3965, followed by 6hrs of treatment with 300ng/ml Pam3CSK, as indicated. *CaMKIIg, Senp3, Ncor* mRNA levels were determined by Q-PCR and normalized to *Gapdh*.